Screening hydrocarbon yields of sunflowers: *Helianthus maximiliani* and *H. nuttallii* (Asteraceae)

Robert P. Adams

Baylor-Utah Lab, Baylor University, 201 N 5500 W, Hurricane, UT, 84737, USA Robert_Adams@baylor.edu

Matt Lavin

Department of Plant Sciences & Plant Pathology, P.O. Box 173150, Bozeman, MT 59717-3150

Steve Hart

Langston University, P.O.Box 730, Langston, OK 73050

Max Licher

P.O. Box 1456, Sedona, AZ 86339

and

Walter Holmes Baylor University, Biology Department, Waco, TX, 76798, USA

ABSTRACT

Populations of perennial *Helianthus maximiliani* Schrad. from eastern Oklahoma, central Texas and Montana were examined for their content of free, extractable hydrocarbons (HC) from leaves. The highest yielding population was at Langston, OK (5.06%), and the lowest yield was in Falls Co, TX (2.2%). The coefficient of variation for % HC yield was 24.3 to 30.0% in central Texas, 26.8% (Langston, OK) and very low, 9.5%, in Bozeman, MT. HC yields (g HC/ g DW plant) were variable in central Texas (0.071 - 0.144 g/ plant), lowest at Bozeman, MT (0.042 g/ plant) and high at Langston, OK (0.098 g/ plant). The percent yields of HC from *H. nuttallii* Torr. & Gray were very high (comparable with the annual, *H. annuus*) in the Glendale, UT population (7.02%), high at Sedona, AZ (5.43%) and low near Kanab, UT (3.12%). The coefficient of variation for % HC yield was highest at Kanab, UT (37.7%), moderate at Glendale, UT (16.4%) and lowest at Sedona, AZ (9.5%). HC yields (g HC/ g DW plant) *H. nuttallii* varied from 0.044 g (Kanab, UT), to 0.088g (Sedona, AZ), to 0.125 g (Glendale, UT). The Glendale, UT population of *H. nuttallii* deserves additional examination due to its high yields. Published on-line www.phytologia.org *Phytologia 100*(2): *161-166 (Jun 22, 2018)*. ISSN 030319430. **KEY WORDS**: *Helianthus maximiliani, H. nuttallii*, Sunflower, yields of hexane soluble leaf hydrocarbon.

Adams and Seiler (1984) surveyed 39 taxa of sunflowers that were grown in a common garden at the USDA lab, Bushland, TX. They analyzed cyclohexane (hydrocarbon), rubber, methanol (resins) yields plus protein concentrations. They reported (Table 1) cyclohexane (HC) yields of 3.10% (IN), and 3.50% (NM) for *H. maximiliani* and 5.25% (NV) and 5.17% (UT) for *H. nuttallii* (Table 1).

Adams et al. (1986) screened 614 taxa from the western US for their hydrocarbon (hexane soluble) and resin (methanol soluble) yields. They reported 4.15% HC from *H. nuttallii*, but they did not examine *H. maximiliani*.

Table 1. Analyses of 39 Taxa of *Helianthus*, representing 49 accessions. Modified from Adams and Seiler, 1984).

Annual (A) or			C-hex	Rubber	MeOH	Total	Percent
Helianthus species peren	nial (P)	Origin	fract.	yield ^a	fract.		Protein ^b
agrestis	А	Brandenton, FL	7.38	1.66	13.45	20.83	6.9
angustifolius	Р	Alvin, TX	3.33	0.18	9.58	12.91	15.9
annuus	А	Winton, OK	7.09	1.40°	11.73	18.82	8.7
annuus, hybrid 894	А	Bushland, TX	2.23	0.49	14.65	16.88	8.6
anomalus	А	Mexican Water, AZ	5.74	0.18	12.30	18.04	9.8
agrophyllus	А	Rockport, TX	6.52	1.14 ^c	9.60	16.12	11.9
arizonensis	Р	Snowflake, AZ	6.13	0.28°	13.16	19.29	18.4
califomicus	Р	Napa, CA	3.05	1.78°	12.44	15.49	13.8
ciliaris	Р	Bushland, TX	5.26	0.57	17.17	22.43	15.6
debilis	А	Titusville, FL	1.95	0.68	8.83	10.78	9.6
deserticola	А	Leeds, UT	3.16	0.82	10.96	14.12	5.3
divaricatus	Р	Wister, OK	1.09	0.47^{d}	11.54	13.44	2.6
glaucophyllus	Р	Blowing Rock, NC	3.29	0.25	9.50	12.79	8.1
grosseserratus	Р	Cherokee Co., KS	2.36	0.28	12.28	14.64	14.6
grosseserratus	Р	Hooker Co., KS	4.41	0.28	14.37	18.78	20.1
grosseserratus	Р	Stuart, OK	3.56	0.28	10.49	14.05	17.1
hirsutus	Р	Wilburton. OK	1.60	0.30	8.30	9.90	6.1
laciniatus	Р	Mimbres River, NM	3.15	0.31	12.40	15.55	9.9
laetiflorus	Р	Lyon Co., KS	2.22	0.66	10.64	12.86	11.9
laevigatus	Р	Botetourt Co., VA	3.53	na	18.24	21.77	13.9
maximiliani	Р	Bloomington, IN	3.10	na	13.21	16.31	10.8
maximiliani	Р	San Jon, NM	3.50	0.24	9.87	13.37	15.3
maximiliani	Р	Gatesville, TX	2.53	na	10.30	12.83	8.9
microcephalus	Р	Cherokee Co., SC	4.77	0.26°	14.25	19.02	14.1
mollis	Р	Greenwood Co., KS	3.26	0.31	11.05	14.31	8.9
mollis	Р	Okmulgee Co., OK	2.60	0.31	9.72	12.32	8.5
mollis	Р	Rivercrest, TX	1.87	0.31	8.58	10.45	6.6
neglectus	Α	Kennit, TX	3.83	0.10	11.71	15.54	16.2
nuttallii	Р	Orovada, NV	5.25	0.96 ^c	10.23	15.48	8.8
nuttallii	Р	Payson, UT	5.17	na	12.76	17.93	10.6
occidentalis	Р	Raymondville, MO	2.12	0.48	15.14	17.26	11.9
occid. ssp. plantagineus	Р	Sheridan, TX	2.36	1.62	18.33	20.69	8.8
paradoxus	А	Ft. Stockton, TX	3.46	0.15	19.54	23.00	13.3
petiolaris ssp.fallax	А	Adrian, TX	2.15	0.30	11.99	14.14	17.3
petiolaris ssp. petiolaris	А	Memphis, TX	1.86	0.14	21.00	22.86	12.1
praecox ssp. hirtus	А	Carrizo Springs, TX	5.19	0.49	10.05	15.24	13.8
pumilis	Р	Boulder, CO	1.72	0.53	6.87	8.59	7.4
resinosus	Р	Collins, MS	2.89	1.78°	11.76	14.65	11.9
rigidus ssp. rigidus	Р	Brookston, IN	1.86	na	9.93	11.79	7.8
rigidus ssp. subrhomboide	s P	Leyden, CO	1.42	na	10.90	12.32	9.9
salicifolius	Р	Kansas	3.13	0.37	9.30	12.43	7.1
salicifolius	Р	Muenster, TX	3.26	0.37	9.31	12.57	11.2
silphioides	Р	Wister, OK	2.63	0.42	18.01	20.64	10.0
simulans	Р	Milton, FL	3.42	0.31	13.91	17.33	18.1
smithii	Р	Morgantown, NC	4.48	0.58°	11.77	16.25	12.2
strumosus	Р	Siler City, NC	2.98	0.55	11.80	14.78	12.9
tuberosus	Р	Kilgore, TX	2.26	0.93	13.28	15.54	12.1
tuberosus x annuus	Р	Turlock, CA	1.73	na	12.21	13.94	9.3
Average			3.39	0.57	12.26	15.65	11.35

a Rubber yields, for leaves except for *divaricatus*, are from Stipanovic *et al* 11 22
b Protein determined by Kjeldahl N x 6-25.
c By 13C·NMR spectral analysis. All others by gravimetric.
d Whole plant analyzed.

Seiler, Carr and Bagby (1991) reported on 28 *Helianthus* taxa for their yields of oil, polyphenols, protein and rubber. The rubber was found to be of lower molecular weight than *Hevea* rubber, but still appeared to be useful as a plasticizing additive and for coatings inside pipes and containers.

Yields of natural rubber has recently been reported for *H. annuus* (Pearson et al. (2010a) that ranged from 0.9% to 1.7% rubber in cultivated sunflowers (Fig. 4, Pearson et al. 2010b).

Most recently, Adams et al. (2018) reported on geographical variation in natural rubber yields in H. *annuus*. They found considerable variation in yields of natural rubber with the highest yielding populations at Mill Creek, UT and in the Waco, TX area. These high yielding populations are in very different eco-systems with different climates and soils. The rubber concentrations in adjacent plants sometimes varied from none to 16 mg/g.

Recently, Adams et al. (2017a) reported on hydrocarbon yields (HC) from a large area from Oklahoma to southern California. This report is a continuation of that survey of HC in sunflowers as we examine HC yields on *Helianthus maximiliani* Schrad. and *H. nuttallii* Torr. & Gray (Adams and TeBeest, 2016; Adams, et al. 2016; Adams et al. 2017b).

MATERIALS AND METHODS

Population locations - see Appendix I.

The lowest growing, non-yellowed, 8 mature leaves were collected at stage R 5.1-5.3 when the first flower head opened with mature rays. The leaves were air dried in paper bags at 49° C in a plant dryer for 24 hr or until 7% moisture was attained.

Leaves were ground in a coffee mill (1mm). 3 g of air dried material (7% moisture) were placed in a 125 ml, screw cap jar with 20 ml hexane, the jar sealed, then placed on an orbital shaker for 18 hr. The hexane soluble extract was decanted through a Whatman paper filter into a pre-weighed aluminum pan and the hexane evaporated on a hot plate (50°C) in a hood. The pan with hydrocarbon extract was weighed and tared. Yields were corrected by a correction factor derived from soxhlet, 6 hr extraction with hexane yields (3) divided by 18 hr shaker with hexane yield(3) = 2.06.

RESULTS

The % HC yields ranged from 2.2 to 5.06% (Table 2) for *H. maximiliani*. The highest yielding population was at Langston, OK (5.06%), and the lowest yield was in Falls Col, TX (2.2%). Yields from populations in central Texas, around Waco, TX, were moderate in % HC yields from 2.20 to 3.68% (Table 2). The Waco, TX yields were comparable to that reported (Adams and Seiler, 1984) (see Table 2, *H. maximiliani*, 3.10%, 3.5%). The population at Langston, OK (5.06%) was higher than previously reported for *H. maximiliani* (Table 1).

The coefficient of variation for % HC yield was 24.3 to 30.0% in central Texas, 26.8% (Langston, OK) and a very low 9.5% in Bozeman, MT (Table 2). The low variation at Bozeman may be due to a small population size.

HC yields (g HC/ g DW plant) were variable (Table 2) in central Texas (0.071 - 0.144 g/ plant), lowest at Bozeman, MT (0.042 g/ plant) and high at Langston, OK (0.098 g/ plant). The low HC g yield was largely due to the very small leaves (1.14 g for 8 leaves). Bozeman is near the northern limit of *H*. *maximiliani*, and, as such, may just produce very small leaves due to the short growing season.

The percent yields of HC from *H. nuttallii* (Table 2) were very high (comparable with the annual, *H. annuus*) in the Glendale, UT population (7.02%), high at Sedona, AZ (5.43%) and low near Kanab, UT (3.12%). The yield from Sedona, AZ (5.43%) is comparable to those reported (Adams and Seiler, 1984) for plants from NV and UT (5.25, 5.17%, respectively, Table 1).

The *H. nuttallii* at the Kanab site has few plants and they grow in the water of a small pond that has afternoon shade and is spring fed in an isolated canyon. This was a very mesic, and lush site that may have resulted in lower amount of stored defense chemicals than in plants less protected (as is the case of the Glendale population).

The coefficient of variation for % HC yield was highest at Kanab, UT (37.7%), moderate at Glendale, UT (16.4%) and lowest at Sedona, AZ (9.5%) (Table 2). The large amount of variation at the Kanab site is surprising (see habitat discussion above). The low amount of variation at the Sedona site may be due to population size.

HC yields (g HC/ g DW plant) *H. nuttallii* varied from 0.044 g, Kanab, UT, to 0.088 g, Sedona, AZ, to 0.125 g, Glendale, UT (Table 2).

The Glendale, UT population of *H. nuttallii* is in a mesic / seasonally dry roadside ditch with grasses and cattails. It extends for about a kilometer and has thousands of plants. This population deserves additional research with seed-grown progeny and transplant studies due to its high HC yields (in progress).

popn id, sample ids	collection number, taxon, location	wt / 8 Ivs	% HC yield	Coef. of var. %	Range of yields	HC g/ plant (8 lvs)
	H. maximiliani					
MxCC	15340, <i>H. maximiliani,</i> Coryell Co., Holmes,	3.40	3.24	30.0	2.17-5.42	0.110
MxFC	15341, <i>H. maximiliani,</i> Falls Co., Holmes,	3.23	2.20	24.3	1.19-3.17	0.071
MxMC	15342, <i>H. maximiliani,</i> McLennan Co., Holmes	3.90	3.68	24.7	2.54-5.42	0.144
MXMT	15276, <i>H. maximiliani,</i> Bozeman Lavin	1.14	3.71	9.5	3.32-4.47	0.042
MXOK	15333, <i>H. maximiliani,</i> Langston, OK, Hart	1.94	5.06	26.8	3.16-7.82	0.098
	H. nuttallii					
NuKUT	15263, <i>H. nuttallii,</i> Kanab, UT Adams	1.40	3.12	37.7	1.7-5.26	0.044
NuGUT	15260, <i>H. nuttallii,</i> Glendale UT Adams	1.78	7.02	16.4	5.58-9.16	0.125
NuSAZ	15290, <i>H. nuttallii,</i> Licher, Sedona, AZ,	1.62	5.43	9.5	4.49-6.07	0.088

Table 2. The yields of hydrocarbons (HC) for H. maximiliani and H. nuttallii.

LITERATURE CITED

- Adams, R. P., M. F. Balandrin, K. J. Brown, G. A. Stone and S. M. Gruel. 1986. Extraction of liquid fuels and chemical from terrestrial higher plants. Part I. Yields from a survey of 614 western United States plant taxa. Biomass 9: 255-292.
- Adams, R. P. and G. J. Seiler. 1984. Whole plant utilization of sunflowers. Biomass 4:69-80.
- Adams, R. P. and A. K. TeBeest. 2016. The effects of gibberellic acid (GA3), Ethrel, seed soaking and pre-treatment storage temperatures on seed germination of *Helianthus annuus* and *H. petiolaris*. Phytologia 98: 213-218.
- Adams, R. P., A. K. TeBeest, W. Holmes, J. A. Bartel, M. Corbet, C. Parker and D. Thornburg. 2017a.
 Geographic variation in pentane extractable hydrocarbons in natural populations of *Helianthus* annuus (Asteraceae, Sunflowers). Phytologia 99: 1-10.
- Adams, R. P., A. K. TeBeest, T. Meyeres and C. Bensch. 2017b. Genetic and environmental influences on the yields of pentane extractable hydrocarbons of *Helianthus annuus* (Asteraceae, Sunflowers). Phytologia 99(2): 186-190.
- Adams, R. P., A. K. TeBeest, S. McNulty, W. H. Holmes, J. A. Bartel, M. Corbet, C. Parker, D. Thornburg and K. Cornish. 2018. Geographic variation in natural rubber yields in natural populations of *Helianthus annuus* (Asteraceae, Sunflowers). Phytologia 100: 19-27.
- Adams, R. P., A. K. TeBeest, B. Vaverka and C. Bensch. 2016. Ontogenetic variation in pentane extractable hydrocarbons from *Helianthus annuus*. Phytologia 98: 290-297.
- Pearson, C. H., K. Cornish, C. M. McMahan, D. J. Rath and M. Whalen. 2010a. Natural rubber quantification in sunflower using automated solvent extractor. Indust. Crops and Prods. 31: 469-475.
- Pearson, C. H., K. Cornish, C. M. McMahan, D. J. Rath, J. L. Brichta and J. E. van Fleet. 2010b. Agronomic and natural rubber characteristics of sunflower as a rubber-producing plant. Indust. Crops and Prods. 31: 481-491.
- Seiler, G. J., M. E. Carr and M. O. Bagby. 1991. Renewables resources from wild sunflowers (Helianthus spp., Asteraceae). Econ. Bot. 45: 4-15.

Appendix I Population locations.

Helianthus maximiliani Schrad.

- MxMT Wet area with cattail and Bebbs willow, near jct. I90 and frontage RD, E of Bozeman. 1-2m tall, common at this site, in wets areas they dry out by middle summer. 45° 40' 29.8" N, 111° 00' 29.4"
 W., 4810 ft., 15 Sept 2017, Gallatin Co., Montana, Coll Matt Lavin sn, Lab Acc. *Robert P. Adams* 15276
- MxMc 1.0 mile southwest of Old Robinson Road, Robinson, 31° 28' 10.69" N, 97° 09' 00.3423" W., 7 Oct 2017, McLennan Co., Texas, Coll: Walter Holmes sn, Lab acc.: *Robert P. Adams 15305*
- MxFC Jct of US Hwy 77 and TX Hwy 7, 1.35 miles northwest of Chilton. 31° 17' 26.30" N, 97° 03' 33.91" W., 9 Oct 2017, Falls Co., Texas, Coll: Walter Holmes sn, Lab acc.: *Robert P. Adams 15306*
- MxCC 12.1 miles east of Gatesville on US Hwy 84, at jct of Tex. Hwy 185, 31° 26' 01.04" N, 97° 32' 24.26" W., 13 Oct 2017, Coryell Co., Texas, Coll: Walter Holmes sn, Lab acc.: *Robert P. Adams* 15309
- MxOK Along dirt road. 35° 53' 10.73" N, 97° 13' 47.21" W. Oct 2017, Logan Co., Oklahoma, Coll: Steve Hart sn, Lab acc.: Robert P. Adams 15310

Helianthus nuttallii Torr. & Gray

- NuGUT Ca. several hundred plants, most flowering now. 3-4 ft tall. in a swale (wet area from excess irrigation water) on the west side of US 89, 2 mi n Glendale, with wild cane, milkweed, in seasonally wet grass ditch. 10 plants sampled for HC. 37° 20' 44" N, 112° 36' 04" W., 5960 ft., 24 Aug 2017, Kane Co., Utah, Coll. *Robert P. Adams 15260*
- NuKUT Ca. 50 plants, most flowering now. 4-5 ft tall. on sandy on edge of perennial pond at Best Friends Center. 10 plants sampled for HC. 37° 08' 22" N, 112° 32' 32.5" W., 5350 ft, 24 Aug 2017, near Kanab, Kane Co., Utah, Coll. *Robert P. Adams 15263*
- NuSAZ Near Indian Gardens, Oak Creek Canyon, n of Sedona. edge of marshy area in outflow form spring, 34° 54' 22.11" N, 111° 43' 35.24" W., 4580 ft., 4 Sept 2017, Coconino Co., Arizona, Coll. Max Licher, sn, Lab Acc. *Robert P. Adams 15290*